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TRITERPENES AND TRITERPENOID GLYCOSIDES FROM THE LEAVES OF *ILEX KUDINCHA*

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Key Word Index—*Ilex kudincha*; Aquifoliaceae; α-kudinlactone; β-kudinlactone; γ-kudinlactone; pomolic acid; triterpenoid glycoside.

Abstract—Two new triterpenes, α- and β-kudinlactone, and six new triterpenoid glycosides, kudinosides D–G, I and J, were isolated from the leaves of *Ilex kudincha*. Their structures have been elucidated by spectroscopic and chemical means.

INTRODUCTION

Ilex kudincha C. J. Tseng is widely used as a traditional beverage in southern China. It is also used in popular medicines and in commercial herbal preparations as a stimulant to the central nervous system, a diuretic, a treatment for sore throats, an aid to losing weight and for the relief of hypertension [1, 2]. Many species belonging to different families and genera [3] are used as its original materials. This paper reports the isolation and elucidation of the structures of two triterpenes (**1** and **2**) and seven triterpenoid glycosides (**4**–**10**).

RESULTS AND DISCUSSION

The ethanol–H_2O (7:1) extract from the leaves of the title plant afforded two new triterpenes (**1** and **2**) and six new triterpenoid glycosides, kudinoside D (**4**), E (**5**), F (**6**), G (**7**), I (**9**) and J (**10**), and a known compound kudinoside H (**8**).

The molecular formula of β-kudinlactone (**1**) was determined as $C_{30}H_{46}O_5$ on the basis of its mass ($[M]^+$ at *m/z* 486) and ^{13}C NMR DEPT spectra. The IR spectrum exhibited absorption bands at 3400 (OH), 1730 (C=O, ester) and 1640 (C=C) cm^{-1}. The ^{13}CNMR DEPT spectrum (Table 1) revealed 30 carbon signals: seven methyls, nine methylenes, four methines and ten quaternary carbons. When β-kudinlactone (**1**) and two known triterpene compounds [4, 5] were compared, it became apparent that β-kudinlactone was an ursolic acid derivative. The signals at δ78.2 (C-3), 66.2 (C-12), 74.4 (C-19) and 85.7 (C-20) were shifted significantly downfield by 20–40 ppm, which indicated that these carbons may be connected with oxygen atoms. C-3 (δ78.2), C-12 (δ66.2) and C-19 (δ74.4) were clearly three alcoholic carbons. The orientation of the hydroxyl groups was shown to be β in the case of C-3 and C-12 and α- in the case of C-19 by means of a NOESY experiment and a consideration of the data in the literature [4, 6, 7]. The C-20 (δ85.7) and C-28 (δ175.4) signals confirmed the presence of a hexacyclic lactone, which the carbonyl absorption band in the IR spectrum (1730 cm^{-1}) strongly suggested had a δ-lactone ring [7–10]. The ^{13}CNMR spectrum indicated two olefinic carbons [δ137.6 (C-18) and 146.6 (C-13)] which were shown to be quaternary carbons by DEPT, and which were ascribed to a tetra substituted double bond. In COLOC experiments, characteristic cross-peaks were observed between the quaternary carbon C-4 and H-23, H-24, H-2 and H-5; between the quaternary carbon C-10 and H-25, H-1, H-5, H-6 and H-9; between the quaternary carbon C-13 and H-12 and H-15; between the quaternary carbon C-18 and H-12 and H-16 and also between C-19 and H-30, and between C-20 and H-19 (Fig. 1). Thus, the signals due to C-13, C-18, C-19 and C-20 were assigned. The data for β-kudinlactone (**1**) compared with that of two known triterpenes, pomolic acid [4] and 27-desoxyphillyrigenin [5], and the results of the detailed analysis of ^1H and ^{13}CNMR spectra with the aid of ^1H–^1HCOSY, ^{13}C–^1HCOSY and COLOC spectra, established that **1** must be 3β,12β,19α-trihydroxyurs-13 (18)-en-28,20β-lactone. The stereostructure of **1** is shown in Fig. 2.

α-Kudinlactone (**2**) was assigned the molecular formula $C_{30}H_{44}O_4$ by mass ($[M]^+$at *m/z* 468) and ^{13}CNMR (Table 1) spectroscopy. The IR spectrum showed absorption bands at 3380 (OH), 1735 (C=O, ester) and 1630 (C=C) cm^{-1}, and ^{13}CNMR-DEPT revealed 30 carbon signals: $CH_3 \times 7$, $CH_2 \times 8$, $CH \times 5$,$C \times 10$. The ^1HNMR spectrum of **2** suggested the presence of a *cis*-disubstituted olefinic proton [δ7.50 (*dd*,

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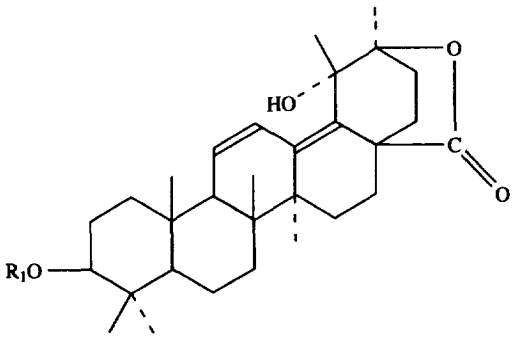
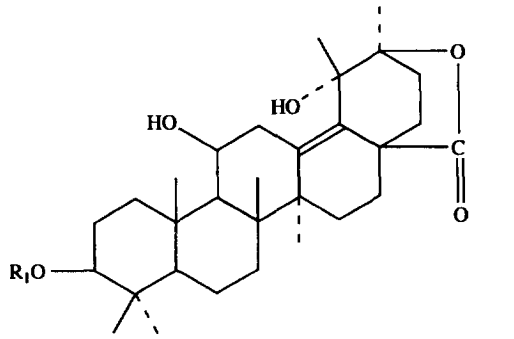
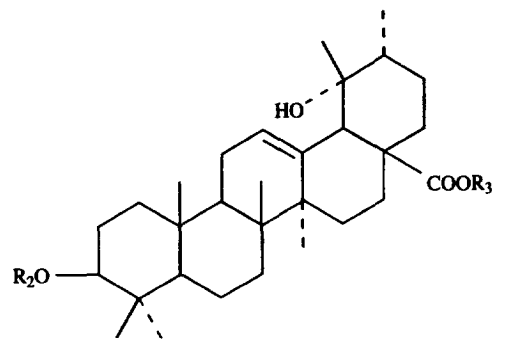


Kudinoside D(4): R_1 = Ara(3)—Glc, Rha(2)—Ara

Kudinoside E(5): R_1 = Ara(3)—Glc(2)—Glc, Rha(2)—Ara



Kudinoside F(6): R_1 = Ara(3)—Glc, Rha(2)—Ara



	R_2	R_3
Kudinoside H(8):	Ara	Glc
Kudinoside G(7):	Ara(3)—Glc, Rha(2)—Ara	Glc

J = 2.8 and 10.2 Hz, H-11) and 5.78 (*d*, J = 10.7 Hz, H-12)]. Comparison of the ^{13}C NMR spectral data for **2** with those of **1** showed that the chemical shifts for C-11, C-12, C-9, C-13, C-25, C-26 and C-27 were changed. The change was due to its stereostructure and conjugated double bond. Thus, **2** was identified as 3β-19α-dihydroxyurs-11(12),13(18)-en-28,20β-lactone.

On acid hydrolysis using 8% H_2SO_4, **9** and **10** furnished β-kudinlactone and α-kudinlactone, respectively, and the same sugar, arabinose. The absolute configuration of the monosaccharide was determined to be L, by HPLC analysis. On comparison of the ^{13}C NMR data for **1** with that for **9**, the downfield shift of the C-3 signal (+ 10.9 ppm) in **9** indicated that the sugar moiety was connected to the C-3 hydroxyl group of β-kudinlactone. Thus, **9** was identified as 3-*O*-α-L-arabinopyranosyl-β-kudinlactone. The corresponding data for **10** showed that this compound was 3-*O*-α-L-arabinopyranosyl-α-kudinlactone.

On negative ion FAB-mass spectroscopy **4** gave a quasi-molecular ion peak $[M]^-$ at *m/z* 908, corresponding to $C_{47}H_{72}O_{17}$. The IR spectrum indicated the presence of a hydroxyl (3400 cm^{-1}) group, a carboxyl (1730 cm^{-1}) group, and a carbon–carbon double bond (1641 cm^{-1}). On cellulase treatment, **4** gave α-kudinlactone as the aglycone ($C_{30}H_{44}O_4$, quasi-molecular ion peak at *m/z* 468 $[M]^+$ in the EI mass spectrum) and a mixture of arabinose, glucose and rhamnose (1:1:1). The absolute configuration of the monosaccharides were determined to be L, D and L, respectively, by HPLC analysis.

In the ^{1}H NMR spectrum (in pyridine-d_5) of **4**, one anomeric proton signal for an L-arabinopyranose moiety was observed at δ4.74 (1H, *d*, J = 5.4 Hz, C-1-H of Ara),

Table 1. ^{13}C NMR chemical shifts of compounds **1**, **2**, **9** and **10** (pyridine-d_5)

	C	1	9	DEPT	2	10	DEPT
Aglycone	1	39.2	39.2	CH_2	38.5	38.1	CH_2
	2	28.3	28.6	CH_2	28.0	28.1	CH_2
	3	78.2	89.1	CH	78.1	88.6	CH
	4	39.5	39.8	C	39.5	39.6	C
	5	56.1	56.3	CH	55.3	55.1	CH
	6	18.7	18.8	CH_2	18.7	18.3	CH_2
	7	35.6	35.8	CH_2	33.1	32.8	CH_2
	8	41.8	42.1	C	42.3	42.2	C
	9	45.0	45.1	CH	54.7	54.4	CH
	10	37.6	37.6	C	37.1	36.6	C
	11	28.9	28.9	CH_2	127.3	127.2	CH
	12	66.2	66.6	CH	128.6	128.4	CH
	13	146.4	146.6	C	140.8	140.7	C
	14	43.9	44.3	C	42.3	42.2	C
	15	28.9	29.3	CH_2	25.9	25.6	CH_2
	16	26.3	27.0	CH_2	26.4	26.1	CH_2
	17	44.1	44.6	C	43.9	43.8	C
	18	137.6	137.6	C	135.2	135.0	C
	19	74.4	74.5	C	74.2	74.1	C
	20	85.7	86.3	C	86.0	85.9	C
	21	28.4	28.6	CH_2	28.6	28.4	CH_2
	22	32.9	32.9	CH_2	33.0	32.8	CH_2
	23	28.8	28.5	CH_3	28.5	27.6	CH_3
	24	16.7	17.2	CH_3	16.1	16.1	CH_3
	25	16.5	16.9	CH_3	18.5	18.3	CH_3
	26	18.3	18.5	CH_3	16.6	16.4	CH_3
	27	23.5	23.8	CH_3	18.7	18.5	CH_3
	28	175.4	176.2	C	175.2	175.2	C
	29	25.3	25.6	CH_3	23.8	23.5	CH_3
	30	19.5	19.8	CH_3	19.6	19.4	CH_3
Sugar	1	C-3-Ara	107.6	CH		107.5	CH
	2		73.1	CH		72.8	CH
	3		74.8	CH		74.5	CH
	4		69.7	CH		69.5	CH
	5		66.8	CH_2		66.7	CH_2

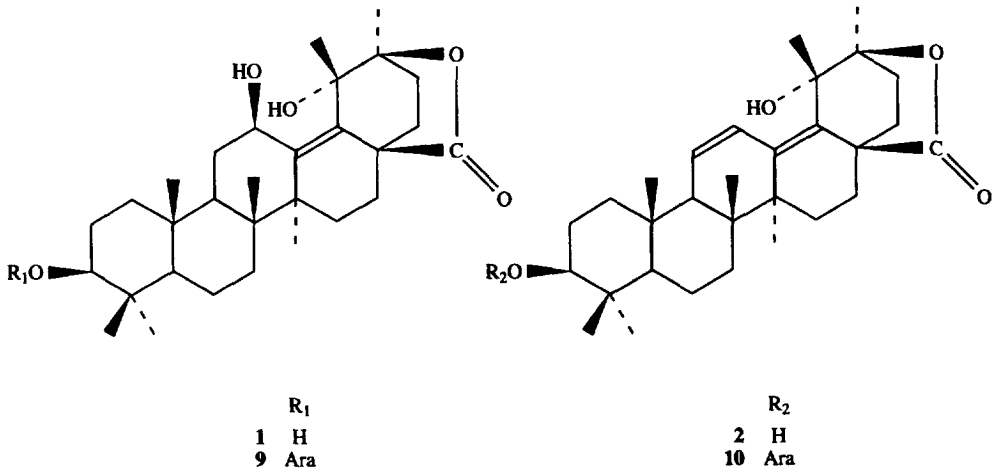
along with the anomeric proton signals for a D-glucopyranose moiety at δ5.14 (1H, *d*, J = 7.7 Hz, C-1-H of Glc) and an L-rhamnopyranose moiety at δ6.40 (1H, *brs*, C-1-H of Rha). Furthermore, in the ^{13}C NMR spectrum, three anomeric carbon signals were observed at δ105.3 (1-C of Ara), 104.9 (1-C of Glc) and 102.0 (J_{C-H} = 170.2 Hz, 1-C of Rha). The J_{C-H} value indicated that the anomeric configuration of the terminal sugar (L-rhamnopyranose) was α [3].

A NOESY experiment on **4** showed the presence of characteristic cross-peaks between signals at δ4.74 (C-1-H of Ara) and 3.34 (C-3-H of aglycone), between δ4.74 (C-1-H of Ara) and 6.40 (C-1-H of Rha), and between δ5.14 (C-1-H of Glc) and 4.23 (C-3-H of Ara).

Based on the foregoing evidence, the chemical structure of **4** has been concluded to be 3-*O*-β-D-glucopyranosyl-(1–3)-[α-L-rhamnopyranosyl (1–2)]-α-L-arabinopyranosyl-α-kudinlactone.

Compound **5** gave a quasi-molecular ion peak $[M-1]^-$ at *m/z* 1069, corresponding to $C_{53}H_{82}O_{22}$, in its negative FAB mass spectrum. The ^1H and ^{13}C NMR spectra showed characteristic signals for an α-kudinlactone glycoside containing arabinopyranose, rhamnopyranose and glucopyranose moieties (Table 2). The IR spectrum showed significant absorption bands due to a hydroxyl (3427 cm^{-1}) group, a carboxyl (1730 cm^{-1}) group and a carbon–carbon double bond (1641 cm^{-1}).

Acid hydrolysis of **5** with 7% H_2SO_4 afforded α-kudinlactone along with L-arabinose, D-glucose and L-rhamnose (1:2:1) as determined by HPLC analysis. Furthermore, in the ^{13}C NMR spectrum glycosylation shifts were observed for the C-2 (Glc) signal (+ 9.2 ppm) and the C-1 (Glc) signal (− 1.6 ppm) of the glucosyl moiety (Glc), demonstrating that the glucopyranosyl group was located at C-2-OH of glucose. It had been presumed that **5** was the C-2 (Glc) glucoside of **4**. Comparison of the sugar moiety of **5** and that of **3** showed the same oligosaccharide sequence. Thus, the chemical structure of **5** was 3-*O*-β-D-glucopyranosyl-(1–2)-β-D-glucopyranosyl-(1–3)-[α-L-rhamnopyranosyl-(1–2)-]-α-L-arabinopyranosyl-α-kudinlactone.



R_1		R_2	
1	H	2	H
9	Ara	10	Ara

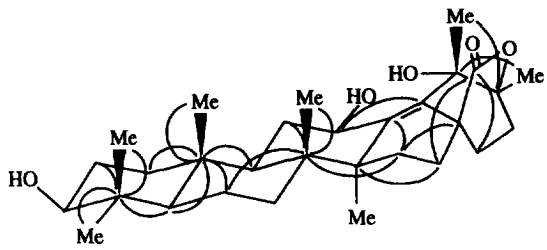


Fig. 1. The results of COLOC for **1**.

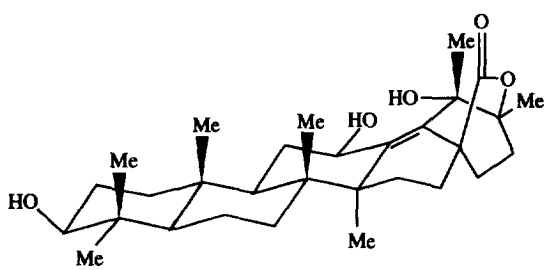


Fig. 2. Stereostructure of **1**.

Compound **6** gave a quasi-molecular ion peak at *m/z* 926 $[M]^-$, which corresponded to $C_{47}H_{74}O_{18}$, in the negative FAB mass spectrum. The IR spectrum showed a similar absorption pattern to that of **4**. On acid hydrolysis with 7% H_2SO_4, **6** provided an aglycone, which was identical to γ-kudinlactone, and a mixture of L-arabinose, D-glucose and L-rhamnose (1:1:1) as determined by HPLC analysis.

By comparison of the ^{13}C NMR data for **6** with that for **4**, it was shown that **6** contains the same oligosaccharide sequence as **4** (Table 2).

A COLOC experiment on **6** exhibited two characteristic cross-peaks between the signal assignable to the proton of the methine at H-11 (δ4.95) and the signal assignable to the quaternary carbons at C-8 (δ43.1) and C-10 (δ37.4). Thus, the aglycone of **6** was the new triterpene, 3β,11β,19α-trihydroxyurs-13(18)-en-28,20β-lactone (γ-kudinlactone). Consequently, the chemical structure of **6** has been determined as 3-*O*-β-D-glucopyranosyl (1–3)-[α-L-rhamnopyranosyl(1–2)-]-α-L-arabinopyranosyl-γ-kudinlactone.

Compound **7** was shown to have the elemental composition of $C_{53}H_{86}O_{22}$ by FAB mass spectroscopy and DEPT. The IR spectrum showed absorption bands due to a hydroxyl (3472 cm^{-1}) group, an ester (1734 cm^{-1}) group and a carbon–carbon double bond (1637 cm^{-1}). On acid hydrolysis with 7% H_2SO_4; **7** provided an aglycone, which was identical to pomolic acid along with a mixture of L-arabinose, D-glucose and L-rhamnose (1:2:1), as determined by HPLC analysis. Alkaline hydrolysis of **7** with aqueous lithium hydroxide afforded compound **11**, which the ^{13}C NMR data showed to contain the same oligosaccharide sequence as **4** or **6** (Table 2), and D-glucose as determined by HPLC analysis, thus demonstrating that a β-D-glucopyranosyl group was located at the C-28 of pomolic acid. The chemical structure of **7** was, therefore, 28-*O*-β-D-glucopyranosyl-pomolic acid 3-*O*-β-D-glucopyranosyl(1–3)-[α-L-rhamnopyranosyl(1–2)-]-α-L-arabinopyranoside.

EXPERIMENTAL

NMR: 400 MHz, pyridine-d_5, chemical shifts (δ) expressed in ppm with solvent as ext. standard. The NMR experiments included ^1H NMR, ^{13}C NMR, ^1H–^1H COSY,

Table 2. ^{13}C NMR data for compounds **4**–**7** (400 MHz, pyridine-d_5)

C	4	5	6	7	Sugar		4	5	6	7
1	38.4	38.5	39.1	39.2	3-Ara	C-1	105.3	105.2	105.2	104.8
2	28.3	28.6	29.5	26.8		2	74.4	74.5	74.0	74.7
3	88.5	88.4	88.9	88.4		3	82.3	82.8	82.5	82.4
4	39.6	39.7	39.9	39.7		4	68.4	69.5	68.3	68.2
5	55.2	55.6	56.3	56.2		5	65.0	65.8	65.1	64.9
6	18.7	18.6	18.6	18.8	Glc	C-1	104.9	103.1	104.9	104.7
7	33.0	33.0	35.5	33.6		2	75.0	84.5	75.1	75.1
8	42.2	42.3	43.1	40.6		3	78.1	78.4	78.2	78.3
9	54.6	54.6	50.6	47.9		4	71.7	71.0	71.6	71.6
10	37.1	37.2	37.7	37.2		5	78.6	78.5	78.7	78.6
11	127.3	127.3	71.5	24.2		6	62.5	62.5	62.5	62.4
12	128.5	128.6	33.7	128.6	GLc	C-1		106.3		
13	140.8	140.9	144.1	139.4		2		76.2		
14	42.2	42.3	45.8	42.2		3		78.3		
15	25.9	26.0	27.5	28.4		4		70.7		
16	26.3	26.4	27.0	26.3		5		78.9		
17	43.8	43.9	46.5	48.8		6		62.1		
18	135.2	135.0	135.7	54.6	Rha	C-1	102.0	101.2	102.1	102.0
19	74.2	74.2	74.3	72.8		2	72.4	72.5	72.4	72.5
20	85.9	86.1	85.8	42.3		3	72.5	72.6	72.5	72.6
21	28.5	28.7	29.5	26.7		4	74.0	74.0	74.1	74.0
22	32.9	33.0	32.8	38.0		5	70.2	69.9	70.5	70.2
23	28.0	27.9	28.4	28.3		6	18.4	18.4	18.8	18.7
24	16.2	16.6	17.3	17.2	28-GLc	C-1				96.0
25	18.4	18.4	17.1	15.9		2				73.7
26	16.6	16.7	18.9	17.5		3				79.0
27	18.7	18.8	22.1	24.7		4				71.4
28	175.4	175.5	176.0	177.3		5				79.3
29	23.7	23.8	27.0	27.2		6				62.6
30	19.4	19.6	20.5	16.8						

^{13}C–^{1}H COSY, DEPT, COLOC and NOESY. IR: KBr discs. The FAB MS: ZAB-HB mass spectrometer; CC: silica gel (60–300 mesh); LPLC: silica gel (10–40 μm); TLC: Silica gel F_{254} coated glass plates using (1) hexane–Me_2CO (2:1) for compounds **1** and **2** and (2) $CHCl_3$–MeOH (10:1) for their glycosides **9** and **10**. Detection: spraying with 10% H_2SO_4 reagent, followed by heating (105°, 5 min), reddish-purple colour.

Plant material. Fresh leaves of *I. kudincha* C. J. Tseng were collected at Yin de, Guangdong Province, China, in July 1992. The plant was identified by Prof. Zi-li Chen. A voucher specimen (No. ICN-34248) is deposited in the Department of Biology, Jinan University. The samples were dried in the dark at room temp. and then coarsely powdered before extraction.

Extraction and isolation. The leaves (5 kg) were extracted (×3) with 70% EtOH (×3) under reflux for 8 hr at 70°. The extract was concd *in vacuo* to yield a dark green residue (500 g) which was suspended in cool water and extracted with Et_2O, EtOAc and *n*-BuOH, respectively. The $CHCl_3$ layer was conc *in vacuo*, and the extract (80 g) subjected to CC on silica gel eluting with hexane (Fr. I) followed by hexane Me_2CO mixts of increasing polarity: 9 : 1 (Fr. II), 4 : 1 (Fr. III), 7 : 3 (Fr. IV), 3 : 2 (Fr. V), 1 : 1 (Fr. VI). Frs IV and V, containing compounds **1** and **2**, over sepd by LPLC (silica gel, 10–40 μm) with hexane–Me_2CO (2 : 1) to give **1** (50 mg) and **2** (20 mg). The EtOAc portion (10 g) was chromatographed on silica gel (200–300 mesh) with $CHCl_3$ and $CHCl_3$–MeOH (20 : 1–10 : 3). The frs were collected. From Frs 8 and 9 [$CHCl_3$–EtOH (10 : 1–5 : 1)] **9** (20 mg) and **10** (15 mg) were obtained.

The *n*-BuOH extract was subjected to TSK gel G 3000S CC (eluting with $H_2O \rightarrow$ MeOH), silica gel CC [eluting with $CHCl_3$–MeOH–H_2O 10 : 3 : 0.5 → $CHCl_3$–MeOH–H_2O 7 : 3 : 0.5 → $CHCl_3$–MeOH–H_2O 65 : 35 : 9), and reversed-phase HPLC (ODS, eluting with MeOH–H_2O 7 : 3 and MeOH–H_2O 9 : 1) to afford **4** (45 mg), E (63 mg), F (35 mg), G (80 mg) and H (22 mg)].

β-*Kudinlactone* (**1**). Powder, molecular formula: 486 ($C_{30}H_{46}O_5$); IR ν_{max}^{KBr} cm^{-1}: 3450–3200 (OH), 1730 (C=O, ester), 1640 (C=C); EIMS *m/z*: 486 $[M]^+$; ^1H NMR (pyridine-d_5) δ (ppm): 0.93 (H-24), 0.93 (H-26), 1.03 (H-25), 1.24 (H-23), 1.50 (H-30), 1.59 (H-27), 1.65 (H-29), 3.45 (1H, *dd*, J = 4.4, 11.2 Hz, H-3), 0.96 (1H, H-5), 2.18 (1H, H-9), 5.92 (1H, H-12); ^{13}C NMR: Table 1.

α-*Kudinlactone* (**2**). Amorphous powder, molecular formula: $C_{30}H_{44}O_4$; EIMS *m/z*: 468 $[M]^+$; ^1H NMR (pyridine-d_5) δ (ppm): 0.86 (H-26), 0.94 (H-25), 1.01 (H-24),

1.03 (H-27), 1.22 (H-23), 1.52 (H-30), 1.68 (H-29), 3.45 (1H, *dd*, $J = 4.2$, 11.5 Hz, H-3), 0.88 (1H, *br s*, H-5), 1.91 (1H, H-9), 7.51 (1H, *dd*, $J = 2.7$, 10.0 Hz, H-11), 5.83 (1H, *d*, $J = 10.5$ Hz, H-12); ^{13}C NMR: Table 1.

Kudinoside (**9**). Amorphous powder; molecular; 618 ($C_{35}H_{54}O_9$); IR ν_{max}^{KBr} cm^{-1}: 3500–3200 (OH), 1728 (C=O, ester), 1639 (C=C); FABMS *m/z*: 625 $[M + Li]^+$, 641 $[M + Na]^+$, 475 $[M + Li - H_2O - \text{pentose}]^+$, 413 $[M + Li - H_2O - \text{pentose} - CO_2]^+$; ^{1}H NMR (pyridine-$d_5$): δ0.89 (H-25), 0.92 (H-26), 0.97 (H-24), 1.29 (H-23), 1.52 (H-30), 1.64 (H-29), 1.65 (H-27), (each 3H, *s*, CH_3), 3.33 (1H, *dd*, $J = 4.02$ Hz, 11.66 Hz, H-3), 0.84 (1H, H-5), 2.19 (1H, *d*, $J = 12.4$ Hz, H-9), 5.96 (1H, *br s*, H-12), 4.78 (1H, *d*, $J = 7.0$ Hz, C-1-H of Ara); ^{13}C NMR: Table 1.

Kudinoside (**10**). Powder, molecular formula: 600 ($C_{35}H_{52}O_8$); FAB-MS *m/z*: 607 $[M + Li]^+$, 613 $[M + 2Li - H]^+$, 563 $[M + Li - CO_2]^+$; ^{1}H NMR (400 MHz, pyridine-d_5): δ0.83 (H-26), 0.89 (H-24), 0.92 (H-25), 1.04 (H-27), 1.15 (H-23), 1.52 (H-30), 1.68 (H-29), 3.34 (1H, *dd*, $J = 4.5$, 11.6 Hz, H-3), 0.86 (1H, H-5), 2.02 (1H, H-9), 7.50 (1H, *dd*, $J = 2.8$, 10.2 Hz, H-11), 5.78 (1H, *d*, $J = 10.7$ Hz, H-12). 4.77 (1H, *d*, $J = 7.0$ Hz, C-1-H of Ara); ^{13}C NMR (400 MHz): Table 1.

Kudinoside D (**4**). Mp 276–279°, $C_{47}H_{72}O_{17}$. IR ν_{max}^{KBr} cm^{-1}: 3500–3100 (OH), 2940 (C-H), 1730 (C=O), 1641 (C=C), 1454, 1360, 1070, 1030; FAB-MS *m/z*: 908 $[M]^-$, 745 $[M - 1 - 162]^-$, 701 $[M - 1 - 162 - CO_2]$, 599 $[M - 1 - 162 - 146]^-$, 555 $[M - 1 - 162 - 146 - CO_2]^-$, 423 $[M - 1 - 162 - 146 - 132 - CO_2]^-$; ^{1}H NMR δ: 0.83 (3H, *s*, CH_3), 0.89 (3H, *s*, CH_3), 0.92 (3H, *s*, CH_3), 1.04 (3H, *s*, CH_3), 1.15 (3H, *s*, CH_3), 1.52 (3H, *s*, CH_3), 1.68 (3H, *s*, CH_3), 3.34 (1H, *dd*, $J = 4.5$, 11.6 Hz, H-3), 0.86 (1H, *br s*, H-5), 2.02 (1H, *br s*, H-9), 7.50 (1H, *dd*, $J = 2.8$, 10.2 Hz, H-11), 5.78 (1H, *d*, $J = 10.7$ Hz, H-12), 4.74 (1H, *d*, $J = 5.4$ Hz, C-1-H of Ara), 5.14 (1H, *d*, $J = 7.7$ Hz, C-1-H of Glc), 6.40 (1H, C-1-H of Rha); ^{13}C NMR: Table 2.

Kudinoside E (**5**). Mp 267–270°, $C_{53}H_{82}O_{22}$. IR ν_{max}^{KBr} cm^{-1}: 3500–3100 (OH), 2940 (C-H), 1730 (C=O), 1641 (C=C), 1450, 1360, 1070; FAB-MS *m/z*: 1069 $[M - 1]^-$, 907 $[M - 1 - 162]^-$, 745 $[M - 1 - 2 \times 162]^-$, 599 $[M - 1 - 2 \times 162 - 146]^-$, 555 $[M - 1 - 162 - 146 - CO_2]^-$, 467 $[M - 1 - 2 \times 162 - 146 - 132]^-$, 423 $[M - 1 - 2 \times 162 - 146 - 132 - CO_2]^-$; ^{1}H NMR δ0.85 (3H, *s*, CH_3), 0.88 (3H, *s*, CH_3), 0.93 (3H, *s*, CH_3), 1.06 (3H, *s*, CH_3), 1.13 (3H, *s*, CH_3), 1.50 (3H, *s*, CH_3), 1.65 (3H, *s*, CH_3), 4.75 (1H, *d*, $J = 7.0$ Hz, C-1-H of Ara), 5.15 (1H, *d*, $J = 7.7$ Hz, C-1-H of Glc), 5.23 (1H, *d*, $J = 7.8$ Hz, C-1-H of GLc), 6.42 (1H, C-1-H of Rha); ^{13}C NMR: Table 2.

Kudinoside F (**6**). Mp 270–274°, $C_{47}H_{74}O_{18}$. IR ν_{max}^{KBr} cm^{-1}: 3450–7200 (OH), 1730 (C=O), 1640 (C=C), 1450, 1380, 1070, 1040; FAB-MS *m/z*: 926 $[M]^-$, 863 $[M - 1 - H_2O - CO_2]^-$, 763 $[M - 1 - 162]^-$, 701 $[M - 1 - H_2O - CO_2 - 162]^-$, 555 $[M - 1 - H_2O - CO_2 - 162 - 146]^-$, 485 $[M - 1 - 162 - 146 - 132]^-$, 423 $[M - 1 - H_2O - CO_2 - 162 - 146 - 132]^-$; ^{1}H NMR δ0.80 (3H, *s*, CH_3), 0.88 (3H, *s*, CH_3), 1.08 (3H, *s*, CH_3), 1.19 (3H, *s*, CH_3), 1.27 (3H, *s*, CH_3), 1.56 (3H, *s*, CH_3), 1.78 (3H, *s*, CH_3), 3.26 (1H, *dd*, $J = 4.2$, 11.3 Hz, H-3), 0.70 (1H, *d*, $J = 10.4$ Hz, H-5), 2.32 (1H, *br s*, H-9), 4.95 (1H, *dd*, $J = 2.3$, 6.4 Hz, H-11), 4.87 (1H, *d*, $J = 5.4$ Hz, C-1-H of Ara), 5.08 (1H, *d*, $J = 7.6$ Hz, C-1-H of Glc), 6.14 (1H, *br s*, C-1-H of Rha), 1.63 (3H, *d*, $J = 5.9$ Hz, C-6-H of Rha); ^{13}C NMR: Table 2.

Kudinoside G (**7**). Mp 228–232°, $C_{53}H_{86}O_{22}$. IR ν_{max}^{KBr} cm^{-1}: 3427 (OH), 2932 (C-H), 1734 (C=O), 1637 (C=C), 1454, 1389, 1072, 1026; FAB-MS *m/z*: 1073 $[M - 1]^-$, 911 $[M - 162]^-$, 765 $[M - 1 - 162 - 146]^-$, 749 $[M - 1 - 2 \times 162]^-$, 603 $[M - 1 - 2 \times 162 - 146]^-$, 453 $[M - 1 - 2 \times 162 - 146 - 132 - H_2O]^-$; ^{1}H NMR δ0.87 (3H, *s*, CH_3), 1.06 (3H, *d*, $J = 6.5$ Hz, CH_3), 1.12 (3H, *s*, CH_3), 1.16 (3H, *s*, CH_3), 1.18 (3H, *s*, CH_3), 1.39 (3H, *s*, CH_3), 1.70 (3H, *s*, CH_3), 3.27 (1H, *dd*, $J = 4.3$, 11.5 Hz, H-3), 4.86 (1H, *d*, $J = 5.4$ Hz, C-1-H of Ara), 5.09 (1H, *d*, $J = 7.7$ Hz, C-1-M of Glc), 6.14 (1H, *br s*, C-1-H of Rha), 1.61 (3H, *d*, $J = 6.1$ Hz, C-6-H of Rha), 6.28 (1H, *d*, $J = 8.0$ Hz, C-1-H of GLc); ^{13}C NMR: Table 2.

Kudinoside H (**8**). Mp 214–215°, $C_{41}H_{66}O_{13}$. IR ν_{max}^{KBr} cm^{-1}: 3400–3100 (OH), 2928 (C-H), 1730 (C=O), 1640 (C=C), 1454, 1380, 1070, 1020; FB-MS *m/z*: 773 $[M + Li]^-$, 789 $[M + Na]^-$, 624 $[M + Li - 1 - 132 - H_2O]^-$, 450 $[M + 2 \times Li - 2 \times H_2O - 162 - 132]^-$; ^{1}H- and ^{13}C NMR: same as zigu-glucoside I [6].

Acid hydrolysis. The sample (**9** or **10**) was dissolved in 8% H_2SO_4 and heated on a water bath at 70° for 8 h. The reaction mixt. was diluted with H_2O and extracted with $CHCl_3$.

Identification of sugar. The water layer was neutralized with 1 N NaOH and concd under red. pres. The residue was compared with standard sugars on TLC ($CHCl_3$–MeOH–H_2O–HOAc, 7 : 3 : 0.5 : 1; detection with spray agent: 4% α-naphthol–EtOH–5% H_2SO_4). The result showed the presence of arabinose.

Acid hydrolysis of kudinosides D, E, F and G. The sample (Kudinoside D, E, F or G) was dissolved in 7% H_2SO_4 (5 ml) and refluxed on a water bath at 90° for 4 hr. The reaction mixt. was diluted with H_2O and extracted with $CHCl_3$. The water layer was neutralized with 1 N NaOH and concd *in vacuo*. The residue was compared with standard sugars on TLC [$CHCl_3$–MeOH–H_2O 7 : 3 : 1, lower layer 9 ml + 1 ml HOAc] and shown to consist of Ara, Glc and Rha each case.

Enzymic hydrolysis of kudinoside D, E, F and G. Kudinoside D (20 mg) or E (25 mg) or C (20 mg) or G (30 mg) was dissolved in EtOH–H_2O (1 : 9) and 0.01 M NaH_2PO_4 buffer (pH 4.0), 5 ml each, and incubated with crude cellulase (50 mg, Sigma) for 2 week at 37°. Usual work up afforded the crude genin which was subjected to CC on silica gel column with C_6H_6–Me_2CO (10 : 1.5). D and E gave α-kudinlactone (15 mg), C gave γ-kudinlactone (6 mg) and G gave pomolic acid. α-Kudinlactone, $C_{30}H_{44}O_4$. IR ν_{max}^{KBr} cm^{-1}: 3300 (OH), 1730 (C=O), 1640 (C=C), 1070, 1030; EIMS *m/z*: 468 $[M]^+$; ^{1}H NMR (pyridine-d_5) δ 0.86 (3H, *s*, CH_3), 0.94 (3H, *s*, CH_3), 1.01 (3H, *s*, CH_3), 1.03 (3H, *s*, CH_3), 1.22 (3H, *s*, CH_3), 1.52 (3H, *s*, CH_3), 1.68 (3H, *s*, CH_3), 3.45 (1H, *dd*,

J = 4.2, 11.5 Hz, H-3), 0.88 (1H, *br s*, H-5), 1.91 (1H, H-9), 7.51 (1H, *dd*, J = 2.7, 10.0 Hz, H-11), 5.83 (1H, *d*, J = 10.5 Hz, H-12).

Alkaline hydrolysis of kudinoside G giving **11**. LiOH (6 mg) was added to a soln of kudinoside G (24 mg) in H_2O (3 ml). The reaction mixt. was heated with stirring at 70° for 10 hr, then cooled to ambient temp., and the solvent removed in a rotary evaporator to give a product (18 mg), which had the same oligosaccharide sequence as **4** or **6**, based on a comparison of the NMR data, and D-glucose, which was determined by TLC analysis.

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